

Amendment under PCT Article 34

45

filed on June 17, 2004

10/511098

DT04 Rec'd PCT/PTO 4 OCT 2004

CLAIMS

1. (Canceled)

5

2. (Canceled)

3. (Canceled)

4. (Canceled)

10

5. (Canceled)

6. (Canceled)

5 7. (Canceled)

8. (Canceled)

9. (Canceled)

10 10. (Canceled)

11. (Canceled)

15 12. (Canceled)

13. (Canceled)

14. (Canceled)

5 15. (Canceled)

16. (Canceled)

10 17. (Canceled)

18. (Canceled)

19. (Canceled)

15 20. (Canceled)

21. (Canceled)

22. (Canceled)

5 23. (Canceled)

24. (Canceled)

10 25. (Canceled)

26. (Canceled)

27. (Canceled)

15 28. (Canceled)

29. (Canceled)

30. (Canceled)

5 31. (Canceled)

32. (Canceled)

33. (Added) An expression vector,  
which comprises: (a) a first coding region encoding  
5 PPIase having molecular chaperone activity, and  
(b) a region having at least one restriction enzyme  
site in which a second coding region encoding a desired  
protein can be inserted.

10 34. (Added) The expression vector according to claim  
33,

wherein the first coding region is operatively  
linked to a promoter, and the restriction enzyme site is  
in the same reading frame as the first coding region, and  
15 is downstream of the first coding region.

35. (Added) The expression vector according to claim  
33 or 34,

20 which has a region being between a first coding  
region and a region having at least one restriction enzyme  
site in which a second coding region can be inserted, and  
is translated in the same reading frame to be a protease  
digestion site.

25 36. (Added) An expression vector,  
wherein a second coding region encoding a desired  
protein is inserted into the expression vector according  
to claim 33, 34 or 35.

30 37. (Added) The expression vector according to claim  
33, 34, 35 or 36,

wherein the PPIase having molecular chaperone  
activity is FKBP-type PPIase.

6 38. (Added) The expression vector according to claim  
33, 34, 35 or 36,

wherein the PPIase having molecular chaperone  
activity is cyclophilin-type PPIase.

5

7 39. (Added) The expression vector according to claim  
33, 34, 35 or 36,

wherein the PPIase having molecular chaperone  
activity is parvulin-type PPIase.

10

4 40. (Added) The expression vector according to claim  
37,

wherein the FKBP-type PPIase is archaeobacterial  
FKBP-type PPIase.

15

9 41. (Added) The expression vector according to claim  
40,

wherein the archaeobacterial FKBP-type PPIase is  
short type FKBP-type PPIase.

20

10 42. (Added) The expression vector according to claim  
33, 34, 35, 36, 37, 38 or 39,

wherein the PPIase having molecular chaperone  
activity comprises an IF domain and/or a C-terminal domain  
of archaeobacterial FKBP-type PPIase.

25

11 43. (Added) The expression vector according to claim  
37,

wherein the FKBP-type PPIase is trigger factor-type  
PPIase.

30

12 44. (Added) The expression vector according to claim  
33, 34, 35, 36, 37, 38 or 39,

wherein the PPIase having molecular chaperone  
activity comprises a N-terminal domain and/or a C-terminal

35

domain of trigger factor-type PPIase.

45. (Added) The expression vector according to claim 37,

5 wherein the FKBP-type PPIase is FkpA-type PPIase.

46. (Added) The expression vector according to claim 33, 34, 35, 36, 37, 38 or 39,

10 wherein the PPIase having molecular chaperone activity comprises a N-terminal domain of FkpA-type PPIase.

47. (Added) The expression vector according to claim 37,

15 wherein the FKBP-type PPIase is FKBP52-type PPIase.

48. (Added) The expression vector according to claim 33, 34, 35, 36, 37, 38 or 39,

20 wherein the PPIase having molecular chaperone activity comprises a C-terminal domain of FKBP52-type PPIase.

49. (Added) The expression vector according to claim 38,

25 wherein the cyclophilin-type PPIase is CyP40-type PPIase.

50. (Added) The expression vector according to claim 33, 34, 35, 36, 37, 38 or 39,

30 wherein the PPIase having molecular chaperone activity comprises a C-terminal domain of CyP40-type PPIase.

51. (Added) The expression vector according to claim 39,

35 wherein the parvulin-type PPIase is SurA-type PPIase.



52. (Added) The expression vector according to claim 33, 34, 35, 36, 37, 38 or 39,

wherein the PPIase having molecular chaperone activity comprises a N-terminal domain of SurA-type PPIase.

53. (Added) The expression vector according to claim 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51 or 52,

wherein the second coding region has a nucleotide sequence encoding a monoclonal antibody.

54. (Added) The expression vector according to claim 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51 or 52,

wherein the second coding region has a nucleotide sequence encoding a membrane protein.

55. (Added) A host, which contains the expression vector according to claim 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53 or 54.

56. (Added) The host according to claim 55, which is *Escherichia coli*.

57. (Added) A fused protein, which comprises PPIase having molecular chaperone activity and a desired protein.

58. (Added) The fused protein according to claim 57, which comprises a protease digestion site between PPIase having molecular chaperone activity and a desired protein.

59. (Added) A process for producing a fused protein comprising PPIase having molecular chaperone activity and a desired protein,

which comprises making the expression vector  
5 according to claim 36, 37, 38, 39, 40, 41, 42, 43, 44, 45,  
46, 47, 48, 49, 50, 51, 52, 53 or 54 express the fused  
protein.

60. (Added) The process for producing a fused  
10 protein according to claim 59,

which comprises culturing the host containing the  
expression vector under condition of expression of the  
expression vector, and making express the fused protein in  
a cytoplasm.

15

61. (Added) The process for producing a fused  
protein according to claim 59,

which comprises providing a region being transcribed  
and translated to be a signal sequence at a 5' terminus of  
20 a first coding region or a 5' terminus of a second coding  
region of the expression vector, and culturing a host  
containing the expression vector under condition of  
expression of the expression vector to express the fused  
protein in a periplasm or a medium.

25

62. (Added) The process for producing a fused  
protein according to claim 59,

which comprises making the expression vector express  
the fused protein in a cell-free translation system.

30

63. (Added) The process for producing a fused  
protein according to claim 59, 60, 61 or 62,

wherein the fused protein is adsorbed on a carrier  
harboring macrolide, cyclosporin, juglone or its analogous  
35 compound inhibiting PPIase activity, and then the carrier

is recovered and the fused protein is recovered from the carrier.

5 64. (Added) A process for producing a desired protein,

which comprises digesting the fused protein comprising a protease digestion site obtained by the process according to claim 59, 60, 61, 62 or 63 with a protease digesting a protease digestion site.

10